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Potential phytotoxic and shading effects of invasive *Fallopia* (Polygonaceae) taxa on the germination of dominant native species

Lenka Moravcová[1], Petr Pyšek[1,2], Vojtěch Jarošík[1,2], Petr Zákravský[1]

1 *Institute of Botany, Academy of Sciences of the Czech Republic, CZ-252 43 Průhonice, Czech Republic*
2 *Department of Ecology, Faculty of Science, Charles University in Prague, Viničná 7, CZ-128 00 Praha 2, Czech Republic*

Corresponding author: *Petr Pyšek* (pysek@ibot.cas.cz)

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Abstract

Two species of the genus *Fallopia* (*F. sachalinensis*, *F. japonica*, Polygonaceae) native to Asia, and their hybrid (*F. ×bohemica*), belong to the most noxious plant invaders in Europe. They impact highly on invaded plant communities, resulting in extremely poor native species richness. The low number of native species in invaded communities points to the possible existence of mechanisms suppressing their germination. In this study we assessed, under laboratory conditions, whether there are phytotoxic effects of the three *Fallopia* congeners on seed germination of three target species: two native species commonly growing in habitats that are often invaded by *Fallopia* taxa (*Urtica dioica*, *Calamagrostis epigejos*), and *Lepidium sativum*, a species commonly used in allelopathic bioassays as a control. Since *Fallopia* taxa form dense stands with high cover, we included varying light conditions as an additional factor, to simulate the effects of shading by leaf canopy on germination. The effects of aqueous extracts (2.5%, 5.0%, and 0% as a control) from dry leaves and rhizomes of the *Fallopia* congeners on germination of the target species were thus studied under two light regimes, simulating full daylight (white light) and light filtered through canopy (green light), and in dark as a control regime. Rhizome extracts did not affect germination. Light treatments yielded inconclusive results, indicating that poor germination and establishment of species in invaded stands is unlikely to be caused by shading alone. However, we found a pronounced phytotoxic effect of leaf extracts of *Fallopia* taxa, more so at 5.0% than 2.5% extract concentration. *Fallopia sachalinensis*

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exerted the largest negative effect on the germination of *Urtica dioica*, *F.* ×*bohemica* on that of *C. epigejos*, and *F. japonica* had invariably the lowest inhibitory effect on all test species. The weak phytotoxic effect of *F. japonica* corresponds to the results of previous studies that found this species to be generally a weaker competitor than its two congeners. Although these results do not necessarily provide direct evidence for allelopathic effects in the field, we demonstrate the potential phytotoxic effect of invasive *Fallopia* taxa on the germination of native species. This suggests that allelopathy may play a role in the impact of *Fallopia* invasion on species diversity of invaded communities.

Keywords
Allelopathy, canopy shading, leaf and rhizome extracts, light regimes, phytotoxicity, plant invasions, *Reynoutria*

Introduction

Recent research on biological invasions increasingly focuses on different types of impacts of invasive species (Levine et al. 2003; Vilà et al. 2010; Pyšek and Richardson 2010), amongst which the impact on the biological diversity of invaded communities and ecosystems is perceived as of utmost importance (Levine 2000; Chornesky and Randall 2003; Sax and Gaines 2003; Richardson and Pyšek 2006; Hejda et al. 2009; Winter et al. 2009). A small fraction of alien invaders change ecological functioning of invaded ecosystems resulting in further changes in the composition and structure of invaded communities (Richardson et al. 2000; Vilà et al. 2010). Invasive plant species have recently been shown to markedly differ in their effects on species richness and diversity of native species in invaded communities; some invaders reduce the numbers of native species that persist after the invasion only to a little extent (Hejda and Pyšek 2006, 2008) while others have considerable impact on native species richness (Hejda et al. 2009). Among the latter group, taxa of the genus *Fallopia* (syn. *Reynoutria*, Polygonaceae) can serve as an example of invasive plants imposing a great impact on native plant species diversity. The stands invaded by *Fallopia* taxa are species-poor, often monospecific (Sukopp and Starfinger 1995; Marigo and Pautou 1998). *Fallopia* taxa exhibit the most severe impacts on species richness and diversity among Central-European alien plants, reducing the number of species present prior to invasion by 66 to 86% (Hejda et al. 2009).

The genus *Fallopia* is native to Asia and several of the taxa are invasive in Europe (Beerling et al. 1994; Bailey and Conolly 2000; Bailey et al. 2007; Lambdon et al. 2008; Pyšek 2009) and other parts of the world (Seiger 1997; Randall 2002; Richards et al. 2008). *Fallopia sachalinensis* (F. Schmidt) Ronse Decr., *F. japonica* (Houtt.) Ronse Decr. var. *japonica* and their hybrid *F.* ×*bohemica* (Chrtek & Chrtková) Bailey, are rhizomatous herbaceous perennials producing a large amount of biomass (Brock 1995; Horn and Prach 1995) and large leaf area (Brabec and Pyšek 2000). They invade along water courses, in waste sites and other disturbed areas (Pyšek et al. 2001, 2002; Pyšek 2009). The invasion of *Fallopia* taxa is among the most intensively studied plant

invasions globally (Pyšek et al. 2008) and has been best documented from the United Kingdom, where it is supposed to have started in Europe (e.g. Bailey et al. 1995; Hollingsworth et al. 1998; Hollingsworth and Bailey 2000; Bailey and Conolly 2000), and from the Czech Republic (Pyšek and Prach 1993; Mandák et al. 2003, 2004).

The ecological understanding of *Fallopia* invasion and impact is still rather poor; it is usually attributed to a high growth rate (Marigo and Pautou 1998), proliferous biomass production (Brock 1995; Horn and Prach 1995), good regeneration ability (Brock et al. 1995; de Waal 2001; Bímová et al. 2003; Pyšek et al. 2003) and its ability to grow at low nutrient levels (Adachi et al. 1996). Possible allelopathic effects of *Fallopia* taxa on germination of other species have not yet been assessed. The large amount of litter produced and the high rhizome density in the soil (Sukopp and Schick 1993) however indicates that this issue may be relevant, considering the low number of species occurring in invaded stands. Allelopathic effects of plant leaf litter were reported as a possible cause of inhibited germination and growth of tree seedlings (Rice 1984) and herbaceous woodland plants (Kuiters and Denneman 1987). Toxic action is attributed to phenolic compounds released from the litter (Rice 1984; Kuiters and Sarink 1986), and *Fallopia* species were reported to contain a large amount of these compounds, e.g. stilbenes, catechins and quinones (Inoue et al. 1992; Vrchotová et al. 2007). Phenolic compounds are known to inhibit not only germination, but also establishment and growth of other species in the community and can also be released into the soil from fresh living tissues such as rhizomes, root bark and leaves (Heisey 1990). In relation to plant invasions, allelopathy has been suggested as one of the underlying mechanisms on which the "novel weapon" hypothesis is based (Callaway and Aschehoug 2000; Hierro and Callaway 2003; Callaway and Ridenour 2004). Many studies have demonstrated the allelopathic effects of invasive plants on native species in their invaded ranges and provided compelling evidence for allelopathy as an important component of the competitive success of some of the world's worst invaders (see Inderjit et al. 2008 and references therein).

The extremely low diversity of *Fallopia*-dominated communities may also be associated with the high cover of *Fallopia* canopy as it has been repeatedly reported that shading by a dense canopy inhibits germination of some plant species (e.g. Górski 1975; Górski et al. 1977; Bewley and Black 1982; Williams 1983; Pons 1992).

To obtain the first insight into the possible role of phytotoxic compounds and shading in reducing diversity of native species present in *Fallopia*-invaded stands, we assessed the effects of these factors on germination of co-occurring native species under laboratory conditions. By using extracts from above-ground and below-ground biomass of two *Fallopia* species and their hybrid, we determined (i) whether the chemical compounds inhibit the germination of two target species, a competitively strong native herb (*Urtica dioica*) and a grass (*Calamagrostis epigejos*), which are often dominant species prior to *Fallopia* invasion. Further, we aimed at assessing (ii) differences in potential phytotoxic action of particular *Fallopia* taxa that are known to differ in their invasiveness, (iii) differences in potential phytotoxic action of leaves and rhizomes, and (iv) explore how these effects are affected by simulated shading.

Material and methods

Target species

Urtica dioica L. is a perennial forb widely distributed in temperate regions (Hultén and Fries 1986). In the Czech Republic, its natural occurrence is in riparian sites where it grows as a dominant species in the summer in floodplain forest understorey. It also occurs as a dominant species of nutrient-rich ruderal habitats (Kopecký and Hejný 1992). In the field, it germinates from March to September. Its fruit (achene) stays dormant for 3–5 months after the collection and dry storage, which enhances germination (Nikolaeva et al. 1985). According to our preliminary tests, 70% of the seeds germinated in 25/10 °C after four months of cold stratification at 6 °C, with more germinating under white light conditions.

Calamagrostis epigejos L. (Roth), a rhizomatous perennial grass of Eurasian distribution (Rothmaler et al. 2002), is one of the most expansive species of the Czech flora, spreading remarkably in the last decades (Prach and Wade 1992). It is typical of disturbed human-made habitats and ruderal grasslands (Kopecký and Hejný 1992) and can grow on extremely acid substrata contaminated by toxic substances (Pyšek and Pyšek 1988). Preliminary tests in 25/10 °C revealed 90% germination after 5 months of dry storage, regardless of light regime. Fruits (caryopses) are produced in large quantities and germinate in the field in spring.

Besides the two native European taxa, an annual forb *Lepidium sativum* L., native to SW Asia and NE Africa (Hultén and Fries 1986) and commonly grown as a vegetable, was used. *Lepidium sativum* is very sensitive to allelopathic compounds, thus often used as a standard target species (Barnes and Putnam 1987; Heisey 1990; Kato-Noguchi and Ino 2001).

Seed and tissue collection

Fruits of target species were collected in Prague, near the Modřanský brook valley (*U. dioica*) and along the road from Modřany to Cholupice village (*C. epigejos*) in September 1999. Seeds of *L. sativum* were obtained from a seed supplier. Fruits of *C. epigejos* and seeds of *L. sativum* were stored in paper bags in the dark at room temperature. Fruits of *U. dioica* were cold-stratified at 6 °C for four months before the start of the experiment.

Fresh rhizomes of the three *Fallopia* taxa were collected in February 1999 in Průhonice near Prague (49°59'41"N, 14°33'56"E). Fresh leaves, without any evident exterior sign of fungal infection, were collected at the same locality in July 1998 and dried at a room temperature.

Extract preparation

Aqueous extracts from dry leaves. Maximum annual dry biomass produced by *Fallopia* in the study area is 228 g/m^2 (P. Pyšek et al., unpublished data). This value was used to set the upper limit of concentrations of phytotoxic compounds used in the experiment. Leaf biomass produced in the field was expressed per area of 75 mm Petri dish used for germination and volume of water it contained (8 ml). This calculation yielded 126 g of dry leaves per litre, i.e. c. 12.5% solution. However, since chemicals are released from leaf tissues gradually, we applied lower concentrations and used 25 and 50 g of dry leaves per litre, respectively, to produce 2.5% and 5% solutions, which is well within the concentration range of allelopathic studies (e.g. Kuiters and Denneman 1987). Dry leaves of *F. sachalinensis*, *F. japonica* and *F. ×bohemica* were ground, weighed and soaked in distilled water. The solutions were left for one week at 20 °C with occasional aeration to prevent anaerobic conditions (Kuiters and Denneman 1987), and then filtered through filter paper. Microbial degradation of the solution was not observed.

Aqueous extracts from rhizomes. Extracts were prepared from fresh washed rhizome cuttings and soaked in 100 ml of distilled water for one week at 20 °C, with occasional aeration to prevent anaerobic conditions. The solutions were filtered through filter paper. Amount of rhizome tissues used corresponded to 10, 20 and 30 g per litre, respectively, yielding 1%, 2% and 3% solution. Microbial degradation of the solution was not observed.

Light regimes applied

Three light regimes were used. Light bulbs and fluorescent tubes provided a white light of 5000 L (105 mmol/cm^2/s^{-1}), simulating daylight in stands not covered by *Fallopia* species. Light filtered through canopy (green light) was simulated by using a single layer of green plastic reducing the red : far-red ratio by 69% and the photon flux density by 56% of white light levels (Skálová and Krahulec 1992); this regime simulated conditions under the *Fallopia* canopy. The standard procedure in germination experiments for creating dark conditions was used, by wrapping Petri dishes with germinating seeds in two layers of aluminium foil.

Seed germination

Germination was tested in growth chambers. Seeds of *L. sativum* (25 seeds), *U. dioica* (50) and *C. epigejos* (50) were placed on two layers of filter paper in 75 mm diameter Petri-dishes and filled with 8 ml of leaf/rhizome extract or distilled water. Filter paper was used as a sterile medium. In the white- and green-light treatments, the evaporated water was replenished where it dropped below a standard mark. Dark treatments

wrapped in aluminium foil could not be filled-up with water during the course of the experiment, but the foil prevented any loss by evaporation. At the end of the experiment we ensured that the petri-dishes did not dry out.

All germination experiments were performed at fluctuating temperature of 25/10 °C (14 h light period/10 h-dark period). Germinating seeds were counted three times a week. Dark treatments were assessed at the end of the experiment, which was terminated after 21 days.

Experimental design

The extracts were first used to conduct preliminary tests on the *L. sativum* seed bioassay. Germination was assessed under controlled conditions (distilled water), two concentrations of dry leaf extract (2.5 and 5%) and three concentrations of fresh rhizome extract (1%, 2% and 3%) of *F. sachalinensis*, *F. japonica* and *F. ×bohemica* with white light. Five replicates were used in each treatment. Since none of the three concentrations of rhizome extracts of the three *Fallopia* taxa had significant effect on germination of *L. sativum*, and seeds of the other two target species germinated up to 100% in most taxon/rhizome extract combinations, fresh rhizome extracts were not used in the main experiment as *L. sativum* is considered to be very sensitive to the allelopathic action.

In the main experiment, we used two concentrations (2.5% and 5%) of leaf extracts from *F. sachalinensis*, *F. japonica* and *F. ×bohemica*, and a control (distilled water = 0%). These combinations were assessed under three light regimes, to test germination response of the three target species, giving the total of 315 Petri dishes (6 taxon/concentration combinations + 1 control × 3 receiver species × 3 light regimes × 5 replicates).

Statistical analysis

Data on the proportion of germinated seeds were angular transformed, and evaluated by fixed effect factorial ANOVAs. The preliminary test was done with a one-way ANOVA, using control and different concentrations of aqueous extracts from leaves and rhizomes of *F. sachalinensis*, *F. japonica* and *F. ×bohemica* as levels of a factor. Differences between control and each extract were assessed by t-tests. The main experiment was evaluated by three-way ANOVA, using the target species (*L. sativum*, *U. dioica* and *C. epigejos*), the light intensity (white, green, dark) and the concentration of aqueous extracts (control, 2.5 and 5% dry leaf extract from *Fallopia sachalinensis*, *F. japonica* and *F. ×bohemica*) as factors. A priori notions on differences among the concentrations within each target species and light intensity were tested by orthogonal contrasts (e.g. Sokal and Rohlf 1994). Calculations were done in the statistical software GLIM® (Crawley 1993l Francis et al. 1994).

Results

The germination of target species varied under the combination of different treatments, from 8.0 to 100.0% in *L. sativum*, from 1.2 to 75.2% in *U. dioica* and from 6.4 to 97.3% in *C. epigejos* (Table 1). Germination was highly significantly affected by the interaction between target species, light regime and leaf extract concentration from the three *Fallopia* taxa (Table 2), and by the interactions between light and leaf extracts within each of the three target species (Table 3 and Fig. 1). In *L. sativum*, the 5% leaf extract from *F. japonica* markedly decreased germination in dark, but not in green and white light; the germination then increased on 2.5% extract from *F. japonica* in dark, but decreased in both light regimes. In *U. dioica*, the decrease in germination between control and 2.5% extract from *F. sachalinensis* was much steeper in light regimes than in dark. In *C. epigejos*, the germination in white light on 2.5% extract from *F. japonica* increased compared to control with no extract; on 5% extract from *F. japonica* germination increased in dark but decrease in both light regimes (Figure 1).

For each target species and light regime, one-way ANOVAs indicated that the germination differed highly significantly among the individual levels of extract concentration from the three *Fallopia* taxa (Table 4). Germination of control seeds also differed highly significantly from that of seeds exposed to extracts, with seed from control samples germinating to higher percentages than those exposed to extracts, except in *U. dioica* in the dark. Thus, *U. dioica* in the dark was the only target species unaffected by phytotoxic effect of extracts.

Within taxa, in all significant cases, 2.5% extracts had lower inhibitory effect than 5% extracts. Extracts from *F. ×bohemica* always produced highly significant differences in germination, and the same was true for extracts from *F. sachalinensis*, except for the effect on *U. dioica* in the white light. *Fallopia japonica* exhibited the weakest difference between concentrations of the extracts: the difference was signifi-

Table 1. Percentage germination of target species under different combination of light treatments (**WL** – white light; **GL** – green light; dark), taxon (**FS** – *Fallopia sachalinensis*; **FJ** – *Fallopia japonica*; **FB** – *Fallopia ×bohemica*) and concentration of the solution (%).

Target species	***Urtica dioica***			***Calamagrostis epigejos***			***Lepidium sativum***		
Treatment	**WL**	**GL**	**Dark**	**WL**	**GL**	**Dark**	**WL**	**GL**	**Dark**
Control	69.6	70.8	20.8	89.6	90.4	97.3	100.0	100.0	100.0
2.5% FS	19.2	28.0	14.8	96.0	71.2	42.4	92.0	86.4	58.4
5% FS	6.8	8.0	1.2	29.6	20.0	12.0	23.2	35.2	8.0
2.5% FJ	73.2	75.2	68.4	96.0	64.0	38.0	96	98.4	100
5% FJ	67.6	51.2	28.8	72.8	20.0	53.6	96.8	96.0	48.8
2.5% FB	71.6	52.4	45.2	86.4	50.4	37.6	93.6	88.0	78.4
5% FB	15.2	28.0	4.8	21.6	6.4	11.2	36.0	59.2	12.0

Table 2. Three-way factorial ANOVA of angular-transformed proportions of germinated seeds for the target species (*Lepidium sativum*, *Urtica dioica* and *Calamagrostis epigejos*), light intensity (white, green, dark) and concentration of aqueous extracts (control, 2.5% and 5% dry leaf extract from *Fallopia sachalinensis*, *F. japonica* and *F. ×bohemica*). The main effects (target species, light and concentration) and first-order interactions (target species) × (concentration), (target species) × (light), and (light) × (concentration), were not tested because the second-order interaction (target species) × (concentration) × (light) was statistically significant.

Source of variation	SS	df	MS	F
Target species	19.25	2	9.625	
Light	3.18	2	1.59	
Concentration	23.17	6	3.862	
(Target species) × (Concentration)	4.57	12	0.381	
(Target species) × (Light)	0.52	4	0.130	
(Light) × (Concentration)	0.80	12	0.0669	
(Target species) × (Concentration) × (Light)	2.01	24	0.08387	4.94 ***
Error	4.28	252	0.01698	
Total	57.78	314		

*** $P < 0.001$

Table 3. Two-way factorial ANOVAs of angular-transformed proportions of germinated seeds for the target species *Lepidium sativum*, *Urtica dioica* and *Calamagrostis epigejos*. The main effects of concentration (control, 2.5% and 5% dry leaf extract from *Fallopia sachalinensis*, *F. japonica* and *F. ×bohemica*) and light (white, green, dark) were not tested because the first-order interaction of the main effects (concentration) × (light) were statistically significant.

Source of variation	*Lepidium sativum*			*Urtica dioica*			*Calamagrostis epigejos*		
	df	MS	F	df	MS	F	df	MS	F
Concentration	6	2.703		6	1.426		6	0.493	
Light	2	0.907		2	0.682		2	0.260	
(Concentration) × (Light)	12	0.142	7.13***	12	0.068	2.74**	12	0.024	3.96***
Error	84	0.0199		84	0.0249		84	0.00613	

*** $P < 0.001$

** $P < 0.01$

cant only in *L. sativum* in dark, in *U. dioica* in green light and in dark, and in *C. epigejos* in white light (Table 4).

Extracts from the three taxa, *F. sachalinensis*, *F. japonica* and *F. ×bohemica*, always significantly differed in their effects on germination (Table 4). Ranked from the strongest to the weakest effect, the extracts from particular *Fallopia* taxa reduced germination in the order *F. sachalinensis* > *F. ×bohemica* > *F. japonica* in *L. sativum* and *U. dioica*, and *F. ×bohemica* > *F. sachalinensis* > *F. japonica* in *C. epigejos*.

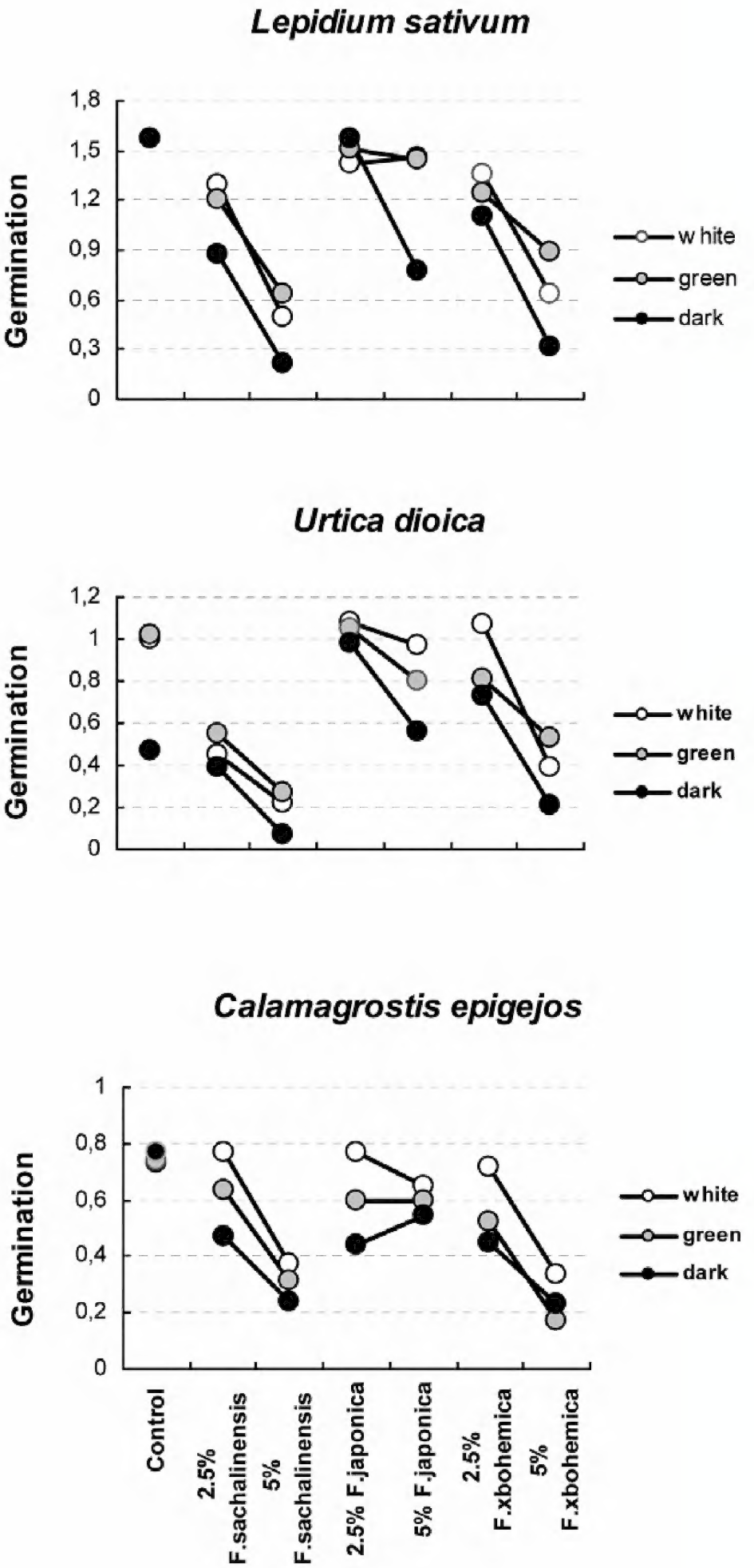

Figure 1. Interactions of angular-transformed proportions of germinated seeds in the target species *Lepidium sativum*, *Urtica dioica* and *Calamagrostis epigejos*. The interactions between the concentration of aqueous extracts (control, 2.5 and 5% dry leaf extract from *Fallopia sachalinensis*, *F. japonica* and *F.* ×*bohemica*) and light intensity (white, green, dark) are apparent as non-parallel lines of light intensity at each panel. See Table 2 for statistical significance of these interactions.

Table 4. One-way ANOVAs of mean angular-transformed proportions of germinated seeds at different concentrations of aqueous extract (control, 2.5% and 5% dry leaf extract from *Fallopia sachalinensis*, *F. japonica* and *F. ×bohemica*) and their orthogonal contrasts (control vs. extracts, *F. sachalinensis* vs. *F. japonica* vs. *F.×bohemica*, and 2.5% vs. 5% dry leaf extract from *Fallopia sachalinensis*, *F. japonica*, and *F. ×bohemica*, respectively) for the target species (*Lepidium sativum*, *Urtica dioica* and *Calamagrostis epigejos*) at different light intensity (white light, green light, dark). Degrees of freedom for ANOVAs among concentrations are df = 6, 28; for the contrast control vs. extracts df = 1, 28; for the contrast *F. sachalinensis* vs. *F. japonica* vs. *F. ×bohemica* df = 2, 28; and for the 2.5% vs. 5% dry leaf extracts df = 1, 28.

Source of variation	*Lepidium sativum*						*Urtica dioica*						*Calamagrostis epigejos*					
	White light		Green light		Dark		White light		Green light		Dark		White light		Green light		Dark	
	SS	MS	SS	MS	SS	MS	SS	MS	SS	MS	SS	MS	SS	MS	SS	MS	SS	MS
Among concentrations	5.489	0.915***	3.576	0.596***	8.863	1.477***	4.139	0.690***	2.375	0.396***	2.860	0.477***	1.044	0.174***	1.182	0.197***	1.025	0.171***
Control vs. extracts	0.919	0.919***	0.746	0.746***	2.495	2.495***	0.401	0.401**	0.527	0.527***	0.0028	0.00280 *ns*	0.0758	0.0758**	0.297	0.297***	0.601	0.601***
F. sachalinensis vs. *F. japonica* vs. *F. ×bohemica*	1.690	0.845***	1.681	0.840***	2.124	1.062***	2.427	1.213***	1.305	0.652***	1.473	0.736***	0.179	0.0896***	0.315	0.157***	0.145	0.0723*
2.5% vs. 5% *F. sachalinensis*	1.608	1.608***	0.826	0.826***	1.087	1.087***	0.135	0.135 *ns*	0.194	0.194**	0.262	0.262***	0.385	0.385***	0.267	0.267***	0.135	0.135**
2.5% vs. 5% *F. japonica*	0.00483	0.00483 *ns*	0.0125	0.0125 *ns*	1.593	1.593***	0.0293	0.0293 *ns*	0.16	0.16**	0.438	0.438***	0.0350	0.0350*	0.00000	0.000000 *ns*	0.0240	0.0240 *ns*
2.5% vs. 5% *F. ×bohemica*	1.267	1.267***	0.311	0.311***	1.564	1.564***	1.146	1.146***	0.188	0.188**	0.685	0.685***	0.369	0.369***	0.303	0.303***	0.120	0.120**
Error		0.0175		0.0205		0.0218		0.0426		0.0183		0.0137		0.00577		0.00531		0.00731

*** P < 0.001, ** P < 0.01, * P < 0.05, *ns* = not significant

Discussion

Potential for allelopathic effects in *Fallopia* taxa

Although the remarkably low species diversity in stands invaded by *Fallopia* is probably due to the competitive advantage resulting from its tall stature, rapid growth, dense rhizome system, and successful regeneration from rhizomes (Brock et al. 1995; Horn and Prach 1995; Bímová et al. 2003; Pyšek et al. 2003), constraints to germination of other species from phytotoxicity and shading may be another contributing mechanisms. The two target species tested, *Urtica dioica* and *Calamagrostis epigejos*, are among the most expansive native species in the Czech Republic (Prach and Wade 1992), dominating in riparian and ruderal habitats where *Fallopia* taxa often invade (Pyšek and Prach 1993; Brabec and Pyšek 2000). Both *C. epigejos* and *U. dioica* are competitively strong rhizomatous perennials forming large stands, but they also produce large quantities of seed (Prach and Wade 1992). In both species, this strategy of regeneration by seed is an effective means of long distance dispersal and colonization of new sites. Attempts to recolonize sites occupied by invasive *Fallopia* taxa might therefore be a common event in the disturbed landscapes where all taxa under question are abundant.

While the effect of light regimes on germination yielded inconclusive results, the interaction of light regime with the inhibitory effect of leaf extracts indicates that poor germination and establishment of native species in stands invaded by *Fallopia* taxa is unlikely to be explained by the effect of shading alone. However, our results demonstrate a pronounced phytotoxic effect of *Fallopia* taxa on germination of native dominant species under laboratory conditions. This inhibition presumably occurred through leaf chemical compounds, was affected by light regime, and its outcome under particular combination of factors depended on the species tested. As pointed out by Baskin and Baskin (1998), light filtered through the canopy can make seeds more sensitive to other environmental factors such as moisture, and thus organic compounds released into the soil could play a role in inhibiting germination until other environmental factors induce dormancy. Phenolic compounds released from leaf litter on the forest floor are permanently present in soil solution but their concentration varies in the course of the vegetation period (Lohdi 1975). Phenology thus plays a role in determining how the possible effect of allelochemicals and shading might operate on a given species. The effect of phenolic compounds is higher in early spring (Lohdi 1975) and for *Fallopia* species, early seasonal development is typical (Marigo and Pautou 1998). In temperate conditions of Central Europe, *Fallopia* quickly builds a dense leaf canopy from April to May (Brabec and Pyšek 2000).

Particular *Fallopia* taxa differed in their effect on germination of the target species. The weakest phytotoxic suppressor was *F. japonica*; its effect on both native dominants was generally lower than that of its two congeners, and higher phytotoxic concentration often did not result in a more pronounced effect. The weak phytotoxic effect of *F. japonica* corresponds to generally lower regeneration (Bímová et al. 2003) and

competitive ability of this species, compared to the other two *Fallopia* taxa (Brabec and Pyšek 2000). The most profound inhibitory effect was found for *F. sachalinensis*, which reduced namely the germination of *U. dioica*. The response of *U. dioica* to *F. ×bohemica* phytotoxins was between that of the two parent *Fallopia* species. Leaf extract of *F. sachalinensis* was previously reported to have other effects, such as stimulation of the synthesis of phenolic compounds in cucumber leaves (Konstantinidou-Doltsinis and Schmitt 1998; Daayf et al. 2000). *Fallopia ×bohemica* also profoundly inhibited germination, particularly that of *C. epigejos*. It may be thus hypothesized that the inhibitory effect of *Fallopia* taxa on germinating native species could be a factor favouring successful invasion.

Limitations of the study

This study was based on using dry leaves, in order to simulate the situation in the field; leaves fall off *Fallopia* plants in dry conditions in the autumn and decomposition proceeds in winter. Although the use of freshly collected and subsequently ground dry leaf material in allelopathical studies has been criticised (Inderjit and Callaway 2003) because green leaves do not have the same properties as true litter and grinding might release compounds that naturally would not leach from leaf cells, this approach is being used to demonstrate the presence of the mechanism (Al Hamdi et al. 2001; Chon et al. 2003; Moradshahi et al. 2003). The present paper aimed at showing whether *Fallopia* above-ground plant parts could potentially inhibit germination of other plant species, and whether this potential differs among the congeners. Given its aim, the study was limited to laboratory conditions, but in order to demonstrate the real significance of the results in invaded communities, further research is needed in the context of soil ecology (Inderjit and Weiner 2001; Hierro and Callaway 2003; Inderjit and Callaway 2003; Inderjit and Nielsen 2003; Inderjit et al. 2008). When interpreting the results, it must therefore be borne in mind that concentrations of phytotoxic allelopathic compounds in the field might be lower than those applied in our study, due to the gradual leaching of compounds from leaves over time and spatio-temporal dynamics of this process (Lohdi 1975).

Our results, nevertheless, demonstrate that there is a strong potential phytotoxic effect of invasive *Fallopia* species on dominant native species and that this effect differs among the three taxa of this genus. Therefore, allelopathy cannot be excluded as one of the mechanisms contributing to the impact on the diversity of native species in *Fallopia*-invaded stands. The results of this paper further indicate that the light regime can influence the outcome of phytotoxic actions and should therefore be taken into account in studies focussing on allelopathic effects of plant species.

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